

Analysis of adaptation mechanism of *Rhodopseudomonas palustris* PSB-S resistance to pyrazosulfuron-ethyl through cytological and proteomic studies

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#Contributed equally to this work.

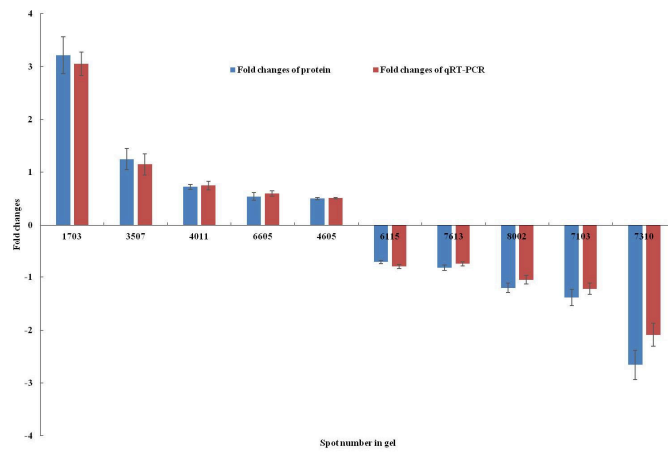
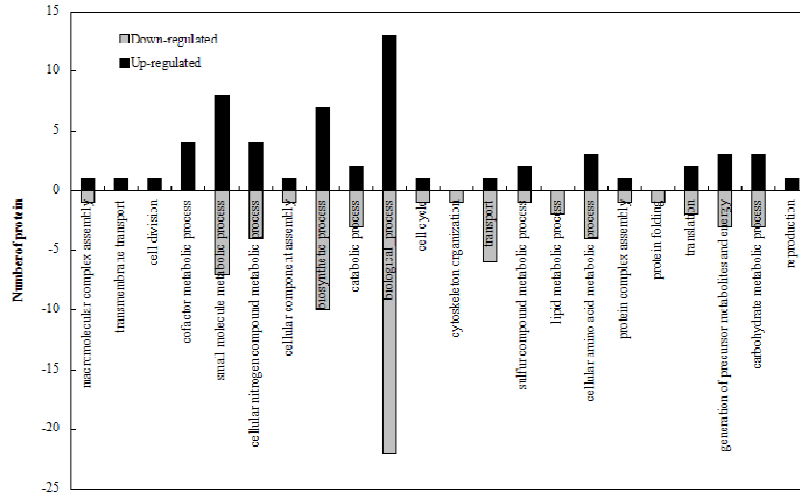
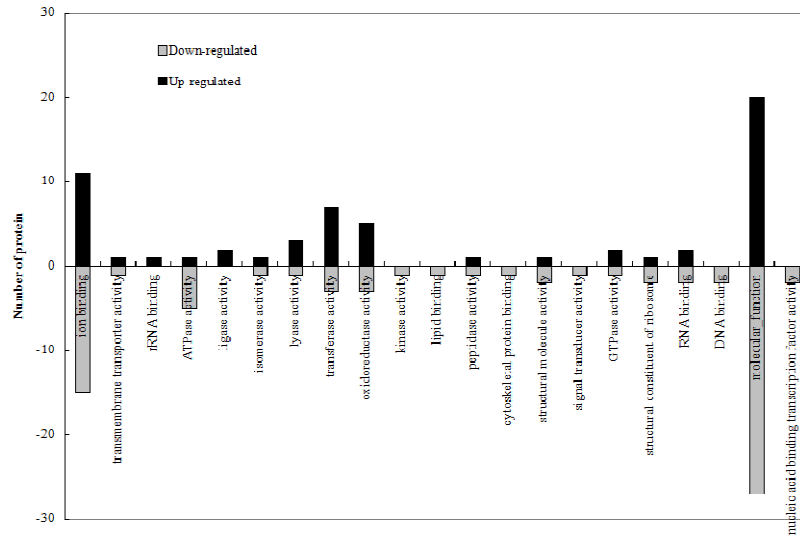


Figure S1. Comparison of results obtained through protein expression analysis (blue bars) or qRT-PCR (red bars). The height of the bars indicate the fold changes. Identification numbers of the analyzed proteins are indicated.

A



B



C

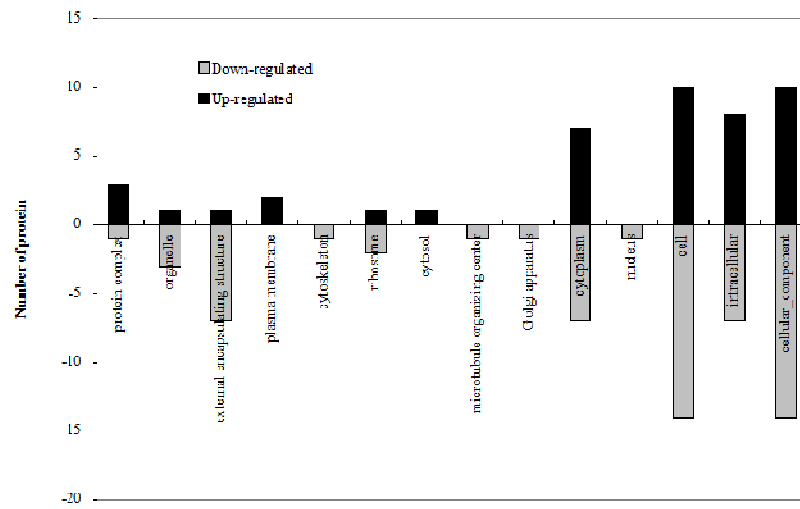


Figure S2. Gene ontology (Go) enrichment of the identified up- or down-regulated proteins in *R. palustris* PSB-S cells treated with 50 µg/ml pyrazosulfuron-ethyl. A protein was considered to be differentially expressed in the pyrazosulfuron-ethyl-treated *R. palustris* PSB-S cells if $t < 0.05$. The GO enrichment analyses were performed using Blast2GO. (A) Number of proteins belonging to various groups in the biological process category. (B) Number of proteins belonging to various groups in the molecular function category. (C) Number of proteins belonging to various groups in the cellular localization category.

Table S1 Differentially expressed proteins during *R.palustris* PSB-S treated with 50 mg/L pyrazosulfuron

Spot no/gel ^a	protein name ^b	Acession number ^c	Ion score ^d	Ion CI ^e	Predict Mv(Da) ^f	predict pI ^g	Log(Fold change) ^h
Upgrade proteins							
1703	Elongation factor G	gi 169830041	73	99.996	76555.51	5	3.21
1604	cell division protein FtsZ	gi 115524129	81	99.994	62562.12	5.13	2.01
3507	ATP synthase subunit alpha	gi 169826598	185	100	54683.81	5.43	1.25
4011	superoxide dismutase	gi 115525836	186	100	22491.03	5.9	0.72
2206	electron transfer flavoprotein subunit alpha	gi 115526836	97	100	32423.13	5.15	0.64
5116	Ribose-5-phosphate isomerase A	sp Q07Q53 RPIA_RHOP5	76	99.999	25256.45	5.51	0.63
5415	O-acetylhomoserine/O-acetylserine sulfhydrylase	gi 91975819	47	92.203	46698.58	6.24	0.61
5607	Succinate dehydrogenase flavoprotein subunit	sp P31038 DHSA_RICPR	54	99.727	65916.47	7.36	0.60
5403	S-adenosylmethionine synthase	sp Q89HP5 METHK_BRAJA	44	97.496	43613.24	5.88	0.57
6605	isocitrate lyase	gi 146340697	71	99.92	61268.64	6.01	0.54
2207	lysine-arginine-ornithine-binding periplasmic protein	gi 90424048	50	93.86	38065.74	8.74	0.53
4605	Acetyl-coenzyme A synthetase	sp Q07VK4 ACSA_RHOP5	43	97.254	72559.08	5.58	0.50
6201	short-chain dehydrogenase/reductase SDR	gi 86750316	160	100	32186.35	6.01	0.47
5504	homospermidine synthase	gi 91975330	72	99.947	53077.3	5.67	0.46
2708	Elongation factor G	sp B3QBY3 EFG_RHOPT	85	100	75837.92	5.27	0.44
3613	chaperonin GroEL	gi 91978588	168	100	57817.73	5.46	0.44
2209	Malate dehydrogenase	sp Q07UX5 MDH_RHOP5	67	99.992	34178.8	5.55	0.44
5313	fructose-1,6-bisphosphate aldolase	gi 148257629	59	99.141	39478.7	5.81	0.44

1516	Aspartyl/glutamyl-tRNA(Asn/Gln) amidotransferase subunit B	sp A9ISC3 GATB_BART1	40	93.25	55467.1	5	0.43
5311	phosphoglycerate kinase	gi 90426241	70	99.945	41864.11	5.95	0.43
5408	S-adenosylmethionine synthetase	gi 115526198	122	100	43207.87	5.88	0.43
3003	30S ribosomal protein S6	sp Q3SR9 RS6_NITWN	59	99.895	15945.16	6.45	0.43
1613	30S ribosomal protein S1	sp Q6NDP1 RS1_RHOPA	76	99.998	62818.75	5.29	0.43
5111	StAR-related lipid transfer protein 9	sp Q9P2P6 STAR9_HUMAN	39	92.166	521839.66	5.91	0.42
6306	fructose-1,6-bisphosphate aldolase	gi 86748075	98	100	39550.75	5.97	0.41
5417	putative leucine/isoleucine/valine-binding protein	gi 91978027	157	100	44363.73	6.38	0.41
Downgrade proteins							
5006	unnamed protein product	gi 27380243	147	100	20377.56	8.86	-0.71
5001	ferric uptake regulator family protein	gi 148251668	80	99.993	17432.8	5.7	-0.71
6107	unnamed protein product	gi 39934274	86	99.998	29441	5.9	-0.76
3604	D-3-phosphoglycerate dehydrogenase	gi 90425581	72	99.96	56105.87	5.87	-0.78
5103	ATP-dependent Clp protease proteolytic subunit	sp A9ISA4 CLPP_BART1	51	99.278	23574.88	5.72	-0.80
8001	histidine triad (HIT) protein	gi 91977514	127	100	15759.1	6.39	-0.80
7003	transcription antitermination protein NusG	gi 115525607	184	100	19932.37	6.36	-0.82
7402	beta alanine--pyruvate transaminase	gi 115522940	62	99.648	48049.44	6.28	-0.82
6111	isopropylmalate isomerase small subunit	gi 91974978	106	100	22495.41	5.29	-0.84
6007	phosphoglyceromutase	gi 115522370	133	100	22939.09	6.32	-0.87
6419	glutamate dehydrogenase, partial	gi 29122978	127	100	38019.55	7.03	-0.87
8113	ABC-type branched-chain amino acid	gi 83311427	62	99.587	25796.56	6.45	-0.89

	transport systems, ATPase component						
6115	ABC transporter-like protein	gi 91977599	78	99.991	27479.41	5.75	-0.89
7214	30S ribosomal protein S2	sp Q215E9 RS2_RHOPB	43.59	95.993	36323.83	6.47	-0.94
7305	branched-chain amino acid ABC transporter	gi 148256992	111	100	45483.64	6.61	-0.94
4009	ubiquinol-cytochrome C chaperone	gi 90424171	66	99.807	20259.22	6.59	-0.99
7104	unnamed protein product	gi 39937216	156	100	25808.4	7.03	-1.02
7308	ABC transporter periplasmic branched chain amino acid binding protein	gi 115526215	152	100	45133.15	7.66	-1.02
7613	acetolactate synthase 3 catalytic subunit	gi 148257335	80	99.994	64316.3	6.2	-1.05
7202	ATPase	gi 146342585	112	100	32307.3	6.14	-1.11
7204	thiosulfate-binding protein	gi 90425493	169	100	36850.11	7.75	-1.11
7313	ABC transporter substrate binding protein	gi 148258710	69	99.935	41990.6	6.62	-1.11
7008	adenine phosphoribosyltransferase	gi 90425859	166	100	21406.32	6.9	-1.11
7107	unnamed protein product	gi 39934752	162	100	22635.76	5.99	-1.14
7603	1-deoxy-D-xylulose-5-phosphate synthase	sp B3QFY7 DXS_RHOPT	128	100	68874.5	6.27	-1.14
8002	acetolactate synthase 3 regulatory subunit	gi 148257333	68	99.928	19991.49	6.43	-1.20
7103	ABC transporter	gi 86749880	111	100	27297.38	6	-1.39
7304	extracellular ligand-binding receptor	gi 86751788	183	100	42536.99	7.67	-1.43
6621	hypothetical protein MT1820.1	gi 15841238	52	96.924	46162.97	6.04	-2.04
7310	ABC-type branched-chain amino acid transport systems periplasmic component	gi 115525850	104	100	48501.43	8.62	-2.66

^aNumber refer to protein designations in Figure5.

^bNames of identified proteins according to the NCBI (<http://www.ncbi.nlm.nih.gov/>) or the Uniprot Knowledgebase (<http://www.uniprot.org>)

^cAccession number according to the NCBI (<http://www.ncbi.nlm.nih.gov/>) or the Uniprot Knowledgebase (<http://www.uniprot.org>)

^dIon score

^eIon C.I.%

^fTheoretical molecular mass (Da), calculated from the corresponding amino acid sequence

^gTheoretical pI, predicted from the corresponding amino acid sequence according to the NCBI (<http://www.ncbi.nlm.nih.gov/>) or the Uniprot Knowledgebase (<http://www.uniprot.org>)

^hProteins differentially expressed by *R.palustris* PSB-S cultivated with 50 mg/L pyrazosulfuron compared with without pyrazosulfuron

Table S2 Primers for qRT-PCR

Spot in gel	Protein name	Prime name	Primer sequence (5'--3')
1703	Elongation factor G	S1F S1R	ATCGGTTCGGAGAACAACCTCA CGGATCAGACGCTTGAGGG
3507	ATP synthase subunit alpha	S2F S2R	AGCCAGAAGCTCTACTGCGTTTA CTGCTTGGACAAGTCGTCATAGA
4011	superoxide dismutase	S3F S3R	GGCTTCGAGAAGTTCAAGACCG TCGACGAACGCCTTCAGGTAG
6605	isocitrate lyase	S4F S4R	CAAGGACGTCAGCAAGTACAACC CGCTGCACGTTCTTAACATAACC
4605	Acetyl-coenzyme A synthase	S5F S5R	GACCGAGTGGCCATCTACAT CAGCCAGCTCCTTCAGGTTCAAA
6115	ABC transporter-like protein	S6F S6R	AGCCGTTCTTTTCCTGCCGTGACC GCCTGCGCCGGCGAAATAGTT
7613	acetolactate synthase 3 catalytic subunit	S7F S7R	CACCGCCTTACCCCTGTTAG ATGCCGTTTCGTCAAATA
8002	acetolactate synthase 3 regulatory subunit	S8F S8R	AAGGGCCGAGAAAAGTGGTT TAGTATAGATCGGGGTGCTG
7103	ABC transporter	S9F S9R	CGCCGCGCCGTTCTT GCAGCGTCGAGGTCTTG
7310	ABC-type branched-chain amino acid	S10F S10R	TCACCGGCGACACCCAGA GCCCCGAACGCCGACG
Reference gene	RubisCO	S11R S11R	ATCCGCAACACGCTGAACGCCGACAA CGACCAGGGTCTTGATCACCGCCAGG